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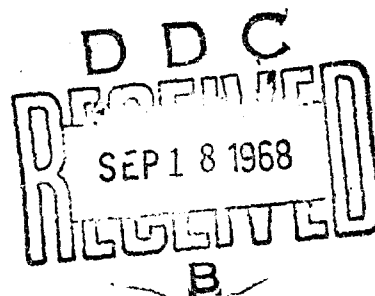
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AD839548

TRANSLATION NO. 1116

DATE: JULY 68



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DEPARTMENT OF THE ARMY
Fort Detrick
Frederick, Maryland

TRANSLATION No. T-181-1 (Blanket Agreement No. CA-18-064-D4-00013(A)
 Bonano, A.M.: Experimental anthrax infection and acidosis and alkalosis
 Experimental anthrax infection and acidotic and alkalotic diet.

Dr. A.M. Bonano - Professor
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 Chairman: Pr. Azzo Azzi

(This report reached the editorial desk on 22 January 1935)

The purpose of this study on the subject of animal immunity is not only the discovery of the innermost nature of the phenomenon, but also the study of the very many conditions which modify the immunity phenomenon in question. The various AA. which have been carried out have controlled from the outside the comportment of the immunity by keeping the animal fasting or by feeding it a special diet: in regards to this, the old experiments of Canalis and Morpurgo (1890) are classical; they concerned the mortality among pigeons, who had been kept without food, of their natural immunity against anthrax or plague. (Gosio and De Giacca).

Thus the aim of this research has been to study the comportment of the immunity process of the animal body, not only when the latter has been kept fasting, but also when it has received insufficient food, quantitatively or qualitatively.

And this series of studies are simulated by the discovery of vitamins, as well as by the recognition of their importance, not only on other vital phenomena, but also on immunity.

We must mention the well-known research carried out by many Italian scientists (Petrognani, Guerrini, Vercellana, Piras and Bruni, A. Ascoli, Setti, etc.) regarding the influence of vitamins on the immunity process, either natural or acquired. I only wish to state that with some AA. the loss of natural immunity in some species of animals who are subjected to a diet devoid of vitamins, there is a decrease in the natural powers of the system; while with other AA., there is an increase in the virulence of the injected germs.

The detection of the phenomenon with a certain degree of probability lies in both factors noted above (decrease in the natural defensive powers and greater virulence of the germs), thus disclosing the intricate relations which regulate the immunity process.

What appears evident from the works cited above is the fact that a non-normal diet as to quantity and quality brings about a disturbance in the extrinsication of the immunity powers of the animal by making it prone to a lesser reactive response against an infective agent, until there is a loss of immunity against a certain infection.

It is also important to note the recent studies of Cormio, in which pigeons and chicken that were kept on a meat diet were found to lose their natural immunity against anthrax.

As it may be seen, the above-mentioned studies are most important: it has occurred to me that it might be worthwhile to expand them by subjecting the test animals to acidotic and alkalotic diets. I was prompted to do so by the fact that, in my previous studies regarding the influence of an acidotic and alkalotic diets on the system, I had done much work regarding the influence of certain of their immunological characteristics and had determined their phagocitory, opsonic, bacteriological, complementary power and their capacity to form anti-bodies and anaphylaxes from the equine serum. I also studied the immunity equilibria and the defense against experimental infection, as well as the comportment of the animal in the case of experimental tuberculosis.

As a conclusion of the above-mentioned studies, I found that the deviations of the acid and base equilibrium, especially in the case of idone, are capable of providing intensive modifications in the immunity equilibrium.

Furthermore, that is obvious, because of the modifications which the above-mentioned diets produce in various organic systems, in the blood texture and in the histological structure of various organs, and because of the complex biochemical modifications of the plasma and of the electronic equilibrium, as that was shown in my research works which I cite above.

Hence the study of the comportment of the natural immunity of animals subjected to an acidotic and alkalotic diet has seemed to me to be a comple-

ment to my previous studies.

Plan of study

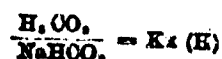
I have chosen the pigeon as the test animal, for that bird is usually immune to anthrax infection.

In order to render virulent the anthrax stock, I followed the method of Patragani and carried out the inoculation to four guinea pigs, by injecting subcutaneously into the abdomen one cubic centimeter of culture in broth. That culture had been made in 24 hours, in a temperature determined by a thermostat, and on the basis of a successively drawn culture of blood, which had itself been drawn from the animal's heart. After the passage of the stock through a rabbit, we obtain a culture in broth that is so virulent that a cubic centimeter of it, when injected subcutaneously into a rabbit weighing 2 kilograms, killed the animal within 48 hours. A normal dose of the film of the culture, which had been made in 24 hours, did not, when diluted in one cubic centimeter of physiological solution and injected into the muscular mass of a pigeon, produce death. That test was repeated with four pigeons weighing an average of 330 grams, thus showing a complete immunity of the animal against anthrax infection.

Then we subjected a group of ten pigeons to an acidotic diet, while an equal number of animals were subjected to an alkalotic diet.

The alkalotic diet pigeons received a food with alkaline ashes (herbs) with a little boiled paste to which we have mixed 1 gr. of sodium carbonate and 50 cg of sodium citrate per pigeon per day; meanwhile, the acidotic diet pigeons received a little paste mixed with a gram of calcium chloride and a gram of ammonium chloride per pigeon per day, as well as small amounts of herbs. Twice a day the animals were forcibly fed, so as to make them eat the amount of meal prepared daily for each group.

Administering such a diet to guinea pigs and rabbits results, as I have shown in my recent works, in a state of acidotic animals and alkalotic animals, the term 'acidotic' indicating synthetically the state of acidity which is non-gaseous and counter-balanced. That is, if following the example of Henderson we indicate the acido-basic equilibrium with a formula that expresses the main regulatory system of such an equilibrium:



we shall consider as a state of acidosis the one ⁱⁿ which the denominator of the equation shown above appears diminished, and in which such a diminution is compensated by a corresponding decrease of the numerator.

On the other hand, we consider as "alkalosis" the state in which we observe a growth of the denominator in question, compensated by a corresponding increase in the numerator.

After 30 days of diet both for the pigeons of the acidosis group and for those of the alkalosis group, they showed a slight change in weight, mainly in the sense of decreased weight.

The injection of a normal dose of a 24 hour culture of anthrax, diluted in one cubic centimeter of physiological solution, carried out in the muscle mass of the hemithrax of certain pigeons, gave the following results:

Acidosis group -

7 pigeons died within 48 hours after the injection.

Alkalosis group -

No pigeon died following the injection of anthrax.

Those results are truly conclusive: the pigeons which had been on an acidosis diet for 30 days lost, in the case of 7 out of a total of 10 of them, their natural immunity against anthrax, but this did not happen to the pigeons which had been kept for a similar period on an alkalosis diet.

Following the example of Petrognani, I wanted to bring about an over-immunization of the pigeons. In order to achieve that, I inoculated the pigeons of the acidosis group, first at the beginning of the diet, and then every 15 days, with virulent anthrax: I found that the injection following the end of the diet produced death through septicemia caused by anthrax, within 52 hours, of eight out of ten pigeons.

Hence the conclusion that an acidosis diet, as well as fasting (Canalis and Morpurgo, Gosio and De Giava), wet cold around the feet (Pasteur), a

diet devoid of vitamins (Petrognani, Piras and Bruni, Vercellana, Guerrini, D'Asaro, Biondo, Ascoli), a meat diet (Cormio) make the pigeon lose its natural immunity against anthrax.

SUMMARY:

An acidotic diet causes the pigeons to lose their natural immunity against anthrax.

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